CLAIMS

1. A method of detecting and/or quantifying an antibody in a liquid sample comprising the steps of

(o') providing a mixture of a liquid phase and a two-component solid phase complex composed of (i) the antibody of the sample, and (ii) a reactant antibody directed against the sample antibody, the reactant antibody being bound to a solid particle,

(p') separating the two-component solid phase complex from the liquid phase,

(q') washing the separated two-component solid phase complex to remove non-complex bound compounds,

(r') adding to the washed two-component solid phase complex a solution of (iii) a ligand in the form of an antigen, an antibody or a hapten, which is optionally labelled, to form a three-component solid phase complex,

(s') optionally adding to the three-component solid phase complex a solution of (iv) a label compound to form a four-component solid phase complex,

(t') separating the three- or four-component solid phase complex obtained in step (r') or (s'), respectively, from the solution,

(u') washing the separated multi-component solid phase complex to remove non-complex bound compounds,

(v') performing a detection/measurement of the washed labelled multi-component complex.

2. A method of detecting and/or quantifying an antibody in a liquid sample comprising the steps of

(o) providing a mixture of a liquid phase and a two-component solid phase complex composed of (i) the antibody of the sample, and (ii) a reactant antibody directed against the sample antibody, the reactant antibody being bound to a solid particle,

(p) separating the two-component solid phase complex from the liquid phase,

(q) washing the separated two-component solid phase complex to remove non-complex bound compounds,

(r) adding to the washed two-component solid phase complex a solution of (iii) a ligand in the form of an antigen, an antibody or a hapten, which is bound to biotin or a functional derivative thereof, to form a three-component solid phase complex,

(s) adding to the three-component solid phase complex a solution of (iv) a chemiluminescent compound covalently bound to avidin, streptavidin or a functional derivative thereof to form a four-component solid phase complex,

(t) separating the four-component solid phase complex from the solution,

(u) washing the separated four-component solid phase complex to remove non-complex bound compound (iv),

(v) initiating a chemiluminescent reaction in the washed four-component solid phase complex and detecting/measuring the resulting chemiluminescence, if any.

3. A method of detecting and/or quantifying an antibody in a liquid sample comprising the steps of

(o'') providing a mixture of a liquid phase and a two-component solid phase complex composed of (i) the antibody of the sample, and (ii) a reactant antibody directed against the sample antibody, the reactant antibody being bound to a solid paramagnetic particle,

(p'') separating magnetically the two-component solid phase complex from the liquid phase,

(q'') washing the separated two-component solid phase complex to remove non-complex bound compounds,

(r'') adding to the washed two-component solid phase complex a solution of (iii) a ligand in the form of an antigen, an antibody or a hapten, which is bound to biotin or a functional derivative thereof, to form a three-component solid phase complex,

(s'') adding to the three-component solid phase complex a solution of (iv) a chemiluminescent compound covalently bound to avidin, streptavidin or a functional derivative thereof to form a four-component solid phase complex,

(t'') separating magnetically the four-component solid phase complex from the solution,

(u'') washing the separated four-component solid phase complex to remove non-complex bound compound (iv),

(v'') initiating a chemiluminescent reaction in the washed four-component solid phase complex and detecting/measuring the resulting chemiluminescence, if any.

4. A method according to claim 2 or 3, c h a r a c t e r i z e d in that the chemiluminescent compound is an acridinium compound.

5. A method according to any of claims 1-3 , c h a r a c t e r i z e d in that component (iii) of step (r'), (r) or (r'') and component (iv) of step (s'), (s) or (s''), respectively, are added in one operation.

6. A method according to any of claims 1-3, c h a r a c t e r i z e d in that the three-component solid phase complex formed in step (r'), (r) or (r'') prior to subjecting it to step (s'), (s) or (s''), respectively, is washed to remove non-complex bound compounds.

7. A method of evaluating and/or predicting the effect of a Specific Allergy Vaccination treatment comprising the steps of

(h') determining the content of an antibody in a liquid sample using the following assay:

(a') providing a mixture of a liquid phase and a three-component solid phase complex composed of (i) the antibody of the sample, (ii) a reactant antibody directed against the sample antibody, the reactant antibody being bound to a solid particle,

and (iii) a ligand in the form of an antigen, an antibody or a hapten,

(b') separating the three-component solid phase complex from the liquid phase,

(c') washing the separated three-component solid phase complex to remove non-complex bound compounds,

(d') adding to the three-component solid phase complex a solution of (iv) a label compound to form a four-component complex,

(e') separating the four-component solid phase complex from the solution,

(f') washing the separated four-component solid phase complex to remove non-complex bound compound (iv),

(g') performing a detection/measurement of the washed labelled four-component complex.

(i') determining the content of the said antibody using the following assay:

(ia') providing a mixture of a liquid phase and a four-component solid phase complex composed of (i) the antibody of the sample, (ii) a reactant antibody directed against the sample antibody, the reactant antibody being bound to a solid particle, (iii) a ligand in the form of an antigen, an antibody or a hapten, and (iv) a label compound, to form a four-component solid phase complex,

(ib') separating the four-component solid phase complex from the liquid phase,

(ic') washing the separated four-component solid phase to remove non-complex bound compounds,

(id') performing a detection/measurement of the washed labelled four-component complex.

(j') comparing the measurements obtained in step (h') and step (i') and using the comparison to evaluate and/or predict the effect of the Specific Allergy Vaccination treatment.

8. A method of evaluating and/or predicting the effect of a Specific Allergy Vaccination treatment comprising the steps of

(h) determining the content of an antibody in a liquid sample using the following assay:

(a) providing a mixture of a liquid phase and a three-component solid phase complex composed of (i) the antibody of the sample, (ii) a reactant antibody directed against the sample antibody, the reactant antibody being bound to a solid particle, and (iii) a ligand in the form of an antigen, an antibody or a hapten, which is bound to biotin or a functional derivative thereof,

(b) separating the three-component solid phase complex from the liquid phase,

(c) washing the separated three-component solid phase complex to remove non-complex bound compounds,

(d) adding to the three-component solid phase complex a solution of (iv) a chemiluminescent compound covalently bound to avidin, streptavidin or a functional derivative thereof to form a four-component solid phase complex,

(e) separating the four-component solid phase complex from the solution,

(f) washing the separated four-component solid phase complex to remove non-complex bound compound (iv),

(g) initiating a chemiluminescent reaction in the washed four-component solid phase complex and detecting/measuring the resulting chemiluminescence, if any.

(i) determining the content of the said antibody using the following assay:

(ia) providing a mixture of a liquid phase and a four-component solid phase complex composed of (i) the antibody of the sample, (ii) a reactant antibody directed against the sample antibody, the reactant antibody being bound to a solid particle, (iii) a ligand in the form of an antigen, an antibody or a hapten, which is bound to biotin or a functional derivative thereof, and (iv) a chemiluminescent compound covalently bound to avidin, streptavidin or a functional derivative

thereof, to form a four-component solid phase complex,

(ib) separating the four-component solid phase complex from the liquid phase,

(ic) washing the separated four-component solid phase to remove non-complex bound compounds,

(id) initiating a chemiluminescent reaction in the washed four-component solid phase complex and measuring the resulting chemiluminescence, if any.

(j) comparing the measurements obtained in step (h) and step (i) and using the comparison to evaluate and/or predict the effect of the Specific Allergy Vaccination treatment.

9. A method of evaluating and/or predicting the effect of a Specific Allergy Vaccination treatment comprising the steps of

(h'') determining the content of an antibody in a liquid sample using the following assay:

(a'') providing a mixture of a liquid phase and a three-component solid phase complex composed of (i) the antibody of the sample, (ii) a reactant antibody directed against the sample antibody, the reactant antibody being bound to a solid paramagnetic particle, and (iii) a ligand in the form of an antigen, an antibody or a hapten, which is bound to biotin or a functional derivative thereof,

(b'') separating magnetically the three-component solid phase complex from the liquid phase,

(c'') washing the separated three-component solid phase complex to remove non-complex bound compounds,

(d'') adding to the three-component solid phase complex a solution of (iv) a chemiluminescent compound covalently bound to avidin, streptavidin or a functional derivative thereof to form a four-component solid phase complex,

(e'') separating magnetically the four-component solid phase complex from the solution,

(f'') washing the separated four-component solid phase complex to remove non-complex bound compound (iv),

(g'') initiating a chemiluminescent reaction in the washed four-component solid phase complex and detecting/measuring the resulting chemiluminescence, if any.

(i'') determining the content of the said antibody using the following assay:

(ia'') providing a mixture of a liquid phase and a four-component solid phase complex composed of (i) the antibody of the sample, (ii) a reactant antibody directed against the sample antibody, the reactant antibody being bound to a solid paramagnetic particle, (iii) a ligand in the form of an antigen, an antibody or a hapten, which is

bound to biotin or a functional derivative thereof, and (iv) a chemiluminescent compound covalently bound to avidin, streptavidin or a functional derivative thereof, to form a four-component solid phase complex,

(ib'') separating magnetically the four-component solid phase complex from the liquid phase,

(ic'') washing the separated four-component solid phase to remove non-complex bound compounds,

(id'') initiating a chemiluminescent reaction in the washed four-component solid phase complex and measuring the resulting chemiluminescence, if any.

(j'') comparing the measurements obtained in step (h'') and step (i'') and using the comparison to evaluate and/or predict the effect of the Specific Allergy Vaccination treatment.

10. A method of evaluating and/or predicting the effect of a Specific Allergy Vaccination treatment comprising the steps of

(x') determining the content of an antibody in a liquid sample using the method of claim 1,

(y') determining the content of the said antibody using the following assay:

(ya') providing a mixture of a liquid phase and a three-component solid phase complex composed of (i) the antibody of the sample, (ii) a reactant

antibody directed against the sample antibody, the reactant antibody being bound to a solid particle, (iii) a ligand in the form of an antigen, an antibody or a hapten, which is labelled or bound to (iv) a label compound, to form a multi-component solid phase complex,

(yb') separating the multi-component solid phase complex from the liquid phase,

(yc') washing the separated multi-component solid phase to remove non-complex bound compounds,

(yd') performing a detection/measurement of the washed labelled multi-component complex.

(z') comparing the measurements obtained in step (x') and step (y') and using the comparison to evaluate and/or predict the effect of the Specific Allergy Vaccination treatment.

11. A method of evaluating and/or predicting the effect of a Specific Allergy Vaccination treatment comprising the steps of

(x) determining the content of an antibody in a liquid sample using the method of claim 2,

(y) determining the content of the said antibody using the following assay:

(ya) providing a mixture of a liquid phase and a four-component solid phase complex composed of (i) the antibody of the sample, (ii) a reactant

antibody directed against the sample antibody, the reactant antibody being bound to a solid particle, (iii) a ligand in the form of an antigen, an antibody or a hapten, which is bound to biotin or a functional derivative thereof, and (iv) a chemiluminescent compound covalently bound to avidin, streptavidin or a functional derivative thereof, to form a four-component solid phase complex,

(yb) separating the four-component solid phase complex from the liquid phase,

(yc) washing the separated four-component solid phase to remove non-complex bound compounds,

(yd) initiating a chemiluminescent reaction in the washed four-component solid phase complex and measuring the resulting chemiluminescence, if any.

(z) comparing the measurements obtained in step (x) and step (y) and using the comparison to evaluate and/or predict the effect of the Specific Allergy Vaccination treatment.

12. A method of evaluating and/or predicting the effect of a Specific Allergy Vaccination treatment comprising the steps of

(x'') determining the content of an antibody in a liquid sample using the method of claim 3,

(y'') determining the content of the said antibody using the following assay:

(ya'') providing a mixture of a liquid phase and a four-component solid phase complex composed of (i) the antibody of the sample, (ii) a reactant antibody directed against the sample antibody, the reactant antibody being bound to a solid paramagnetic particle, (iii) a ligand in the form of an antigen, an antibody or a hapten, which is bound to biotin or a functional derivative thereof, and (iv) a chemiluminescent compound covalently bound to avidin, streptavidin or a functional derivative thereof, to form a four-component solid phase complex,

(yb'') separating magnetically the four-component solid phase complex from the liquid phase,

(yc'') washing the separated four-component solid phase to remove non-complex bound compounds,

(yd'') initiating a chemiluminescent reaction in the washed four-component solid phase complex and measuring the resulting chemiluminescence, if any.

(z'') comparing the measurements obtained in step (x'') and step (y'') and using the comparison to evaluate and/or predict the effect of the Specific Allergy Vaccination treatment.

13. A method according to claim 7, 8, 9, 10, 11 or 12, wherein step (ia'), (ia), (ia''), (ya'), (ya) or (ya''), respectively, is carried out by mixing components (i) and (ii), then adding component (iii), and finally adding component (iv), if added.

14. A method according to claim 7, 8, 9, 10, 11 or 12, wherein step (ia'), (ia), (ia''), (ya'), (ya) or (ya''), respectively, is carried out by mixing components (i), (ii) and (iii), and then adding component (iv), if added.

15. A method according to claim 7, 8, 9, 10, 11 or 12, wherein the comparison of step (j'), (j), (j''), (z'), (z) or (z''), respectively, is carried out by calculating the ratio of the measurements obtained in the two said steps.

16. A method according to any of claim 7, 8, 9, 10, 11 or 12, wherein the comparison of step (j'), (j), (j''), (z'), (z) or (z''), respectively, is carried out at a number of points in time at the start of and during the treatment period, and that any temporal change, which may be observed, is used as a basis for evaluating and/or predicting the effect of the treatment.

17. A method according to claim 1, c h a r a c t e r i z e d in that the label compound is a luminescent label, a chemiluminescent label, an enzyme label, a radioactivity label, a fluorescent label or an absorbance label.

18. A method according to claim 1, c h a r a c t e r i z e d in that the labelled ligand is labelled by a radioactive atom.

19. A method according to claim 1, c h a r a c t e r i z e d in that the separation of the solid phase complex from the liquid phase is carried out by magnetic separation, filtration, sedimentation, centrifugation, chromatography or column chromatography.

20. A method of evaluating the immunological status of a subject comprising the steps of

1) determining the content of an antibody in a liquid sample from the subject using an immunoassay, wherein the reaction between the antibody of the sample and a ligand in the form of an antigen, an antibody or a hapten, the ligand being directed to the Fab region of the sample antibody, is carried out in the presence of other constituents of the sample to obtain a measurement 1,

2) determining the content of an antibody in the liquid sample using an immunoassay, wherein the reaction between the antibody of the sample and a ligand in the form of an antigen, an antibody or a hapten, the ligand being directed to the Fab region of the sample antibody, is carried out in the absence of other constituents of the sample to obtain a measurement 2,

3) interrelating measurements 1 and 2 to express the interference and using the interference as a parameter for evaluating the immunological status of the subject.

21. A method of evaluating the immunological status of a subject comprising the steps of

A) determining the content of an antibody in a liquid sample from the subject using the following assay protocol (assay A):

(Aa) mixing (i) the antibody of the sample, (ii) an antibody directed against the Fc region of the sample antibody, the reactant antibody being bound to a solid carrier and (iii) a ligand in the form of an antigen, an

antibody or a hapten, the ligand being directed to the Fab region of the sample antibody, to form a mixture of a three-component solid phase complex and a liquid phase,

(Ab) contacting the three-component complex with (iv) a label compound to form a mixture of a four-component complex and a liquid phase,

(Ac) washing the four-component solid phase to remove non-complex bound compounds,

(Ad) performing a detection/measurement of the washed labelled four-component complex to obtain a measurement A;

B) determining the content of the said antibody in the said sample using the following assay protocol (assay B):

(Ba) mixing (i) the antibody of the sample, and (ii) a reactant antibody directed against the Fc region of the sample antibody, the reactant antibody being bound to a solid carrier, to form a mixture of a two-component solid phase complex and a liquid phase,

(Bb) washing the two-component solid phase complex to remove non-complex bound compounds,

(Bc) contacting the washed two-component solid phase complex with a (iii) a ligand in the form of an antigen, an antibody or a hapten, the ligand being bound to the Fab region of the sample antibody, and (iv) a label compound, to form a mixture of a four-component solid phase complex and a liquid phase,

(Bd) washing the four-component solid phase complex to remove non-complex bound compounds,

(Be) performing a detection/measurement of the washed labelled four-component complex to obtain a measurement B; and

(E) interrelating measurements A and B to express the interference and using the interference as a parameter for evaluating the immunological status of the subject.

22. A method according to claim 21, wherein step Ab is effected by adding said (iv) label compound to said mixture of a three-component solid phase complex and a liquid phase in step Aa.

23. A method according to claim 21, wherein step Ab is effected by washing the three-component complex obtained in step Aa to remove non-complex bound compounds and by subsequently adding said (iv) label compound to the washed three-component complex.

24. A method according to any of claims 21-23, wherein step Bd is effected by simultaneous incubation of said (iii) ligand and said (iv) label compound with the two-component complex.

25. A method according to any of claims 21-23, wherein initially said (iii) ligand is added to the two-component complex to form a three-component complex, which is then washed to remove non-complex bound (iii) ligand, and then said (iv) label compound is added to form a four-component complex.

26. A method according to any of claims 20-25, wherein the label compound is a luminescent label, a chemiluminescent label, an enzyme label, a radioactivity label, a fluorescent label or an absorbance label.

27. A method according to any of claims 20-26, wherein the (iii) ligand is biotinylated.

28. A method according to claim 27, wherein the (iv) label compound is a chemiluminescent compound covalently bound to avidin, streptavidin or a functional derivative thereof.

29. A method of evaluating the immunological status of a subject comprising the steps of

C) determining the content of an antibody in a liquid sample from the subject using the following assay protocol (assay C):

(Ca) mixing (i) the antibody of the sample, (ii) an antibody directed against the Fc region of the sample antibody, the reactant antibody being bound to a solid carrier and (iii) a labelled ligand in the form of an antigen, an antibody or a hapten, the ligand being directed to the Fab region of the sample antibody, to form a mixture of a three-component solid phase complex and a liquid phase,

(Cb) washing the three-component solid phase to remove non-complex bound compounds,

(Cc) performing a detection/measurement of the washed labelled four-component complex to obtain a measurement C;

D) determining the content of the said antibody using the following assay protocol (assay D):

(Da) mixing (i) the antibody of the sample, and (ii) a reactant antibody directed against the Fc region of the sample antibody, the reactant antibody being bound to a solid carrier, to form a mixture of a two-component solid phase complex and a liquid phase,

(Db) washing the two-component solid phase complex to remove non-complex bound compounds,

(Dc) contacting the washed two-component solid phase complex with a (iii) a labelled ligand in the form of an antigen, an antibody or a hapten, the ligand being bound to the Fab region of the sample antibody, to form a mixture of a three-component solid phase complex and a liquid phase,

(Dd) washing the three-component solid phase complex to remove non-complex bound compounds,

(De) performing a detection/measurement of the washed labelled three-component complex to obtain a measurement D; and

(E) interrelating measurements C and D to express the interference and using the interference as a parameter for evaluating the immunological status of the subject.

30. A method according to claim 29, wherein the labelled ligand is labelled by a radioactive atom.

31. A method according to any of claims 20-30, wherein the subject to be evaluated is undergoing allergy treatment, allergy vaccination treatment or Specific Allergy Vaccination (SAV) treatment.

32. A method of evaluating the effect of allergy treatment of a subject comprising the steps of

A) determining the content of the said antibody using the following assay protocol (assay A):

(Aa) mixing (i) the antibody of the sample, (ii) an antibody directed against the Fc region of the sample antibody, the reactant antibody being bound to a solid carrier and (iii) a ligand in the form of an antigen, an antibody or a hapten, the ligand being directed to the Fab region of the sample antibody, to form a mixture of a three-component solid phase complex and a liquid phase,

(Ab) contacting the three-component complex with (iv) a label compound to form a mixture of a four-component complex and a liquid phase,

(Ac) washing the four-component solid phase to remove non-complex bound compounds,

(Ad) performing a detection/measurement of the washed labelled four-component complex to obtain a measurement A;

(E) using measurement A as a parameter for evaluating the effect of the treatment.

33. A method of evaluating the effect of allergy treatment of a subject comprising the steps of

(C) determining the content of the said antibody using the following assay protocol (assay C):

(Ca) mixing (i) the antibody of the sample, (ii) an antibody directed against the Fc region of the sample antibody, the reactant antibody being bound to a solid carrier and (iii) a labelled ligand in the form of an antigen, an antibody or a hapten, the ligand being directed to the Fab region of the sample antibody, to form a mixture of a three-component solid phase complex and a liquid phase,

(Cb) washing the three-component solid phase to remove non-complex bound compounds,

(Cc) performing a detection/measurement of the washed labelled four-component complex to obtain a measurement C;

(E) using measurement C as a parameter for evaluating the effect of the treatment.

34. A method according to claim 32 or 33, wherein the subject to be evaluated is undergoing allergy vaccination treatment or Specific Allergy Vaccination (SAV) treatment.

35. A method according to any of claims 31-34, wherein the evaluation in step E) is carried out at a number of points in time at the start of and during the treatment period, and that any temporal change, which may be observed, is used as a basis for evaluating and/or predicting the effect of the treatment.

~~38~~ 4. A method according to any of claims 20-34, wherein the carrier is a particle.

~~36~~ 7. A method according to claim 35, wherein the carrier is a paramagnetic particle.

~~37~~ 8. A method according to any of claims 20-36, wherein the sample antibody is a specific IgE.

Rule 1.126

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Add A20